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JIS

JAPANESE INDUSTRIAL STANDARD

Methods for quantitative analysis of lipase

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Methods for quantitative analysis of lipase K 0608-1996

1. Scope This Japanese Industrial Standard specifies the methods for quantitative analysis to determine the quality of lipase to be used for chemical products or for reagent.

Remarks 1. The standards cited in this Standard are listed in Attached Table 1.

2. The classified number of lipase according to the Enzyme Committee of International Union of Biochemistry is EC 3.1.1.3.

2. Definitions The definitions of terms used in this Standard shall follow JIS K 0211 and JIS K 3600.

3. Matters in common The general matters commonly used in chemical analysis shall follow JIS K 0050. Water shall be, however, A3 or A4 specified in JIS K 0557.

4. Classification of quantitative analyses The quantitative analyses for lipase shall be classified into an overall-determination type absorptiometry (Lowry process), a fractional-determination type high performance liquid chromatograph-ultraviolet absorptiometry, and a determination method using enzyme activity.

(1) Absorptiometry (Lowry process) Carry out determination using the absorbance caused by protein-copper complex which was produced by the reaction between protein and copper ion, and phosphomolybdic blue and phosphotungstic blue produced by being reduced with protein. This method can overall determine the protein from 100 to 1 000 mg/*l*.

Remarks: If sample contains 2.5 mmol/*l* or more dihydrogen disodium ethylenediaminetetraacetate, 15 mmol/*l* or more 2-mercaptoethanol, or 0.5 mol/*l* or more sodium dodecyl sulfate, the sample cannot be determined.

(2) High performance liquid chromatograph-ultraviolet absorptiometry Fractionate protein using a high performance liquid chromatography, and determine it with wavelength of near 220 nm. This method can fractionate contained ingredients depending on its molecular size, and determine the lipase from 20 to 100 mg/*l*.

Remarks: If sample contains 0.2 mol/*l* or more dihydrogen disodium ethylenediaminetetraacetate, 50 mmol/*l* or more 2-mercaptoethanol, or 3 mmol/*l* or more sodium dodecyl sulfate, the sample cannot be determined.

(3) Determination method using enzyme activity Under specified temperature and pH condition, let lipase react upon substrate, measure the concentration of the substance which has been generated, and determine the lipase. This method can determine the lipase from 4 to 20 mg/*l*.

Remarks: If it contains the substance likely affecting the activity of enzyme, it cannot be determined.